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Alan Israel
Pacific University

Jill Habib
Pacific University

Larry Danson
Pacific University

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FLUORESCEIN ANGIOGRAPHY

By

ALAN ISRAEL
JILL HABIB
LARRY DANSON

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Advisor:

Salisa K. Williams, O.D.

Alan Israel

Jill Habib

Larry Danson

Salisa K. Williams, O.D.

Alan Israel was born and raised in the Seattle area. He graduated Mercer Island High School in 1985. Alan completed his bachelor of science degree in Visual Science at Pacific University in 1992. After receiving his doctoral degree at Pacific University, he plans on settling in the Seattle area with Jill Habib, whom he will marry in June, 1994.

Jill Habib was born and raised in Fresno, California. She graduated from Washington Union High School. In 1992, she completed her bachelors of science degree in Visual Science at Pacific University. After receiving her doctoral degree at Pacific University, she plans on settling in the Seattle area with Alan Israel, whom she will marry in June, 1994.

Larry Danson was born and raised in Phoenix, Arizona. In 1992, he completed his Bachelor of Science degree in Visual Science at Pacific University. After receiving his doctoral degree at Pacific University, he plans on settling in the Phoenix area with his wife, Paula.

Abstract

Fluorescein angiography is an important diagnostic test used for differential diagnosis of retinal disease. It is important for the optometric physician to have a working knowledge of the anatomy, pharmacology, and the mechanics of this diagnostic procedure. The purpose of this paper is to aid the optometric physician in understanding the basic techniques in performing fluorescein angiography.

(Key words: fluorescein, hypofluorescence, hyperfluorescence, RPE (retinal pigmented epithelium), retina)

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FLUORESCEIN ANGIOGRAPHY

(Prepared by: Alan Israel, Jill Habib, & Larry Danson)
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Description

Fluorescein angiography (FA) is a diagnostic photography procedure used to detect vascular compromise to the retina, and optic nerve. It may also be used to identify areas of the fundus amenable to laser treatment, and to evaluate and monitor postlaser success. FA studies the presence and extent of intra, extra, and subretinal vasculature alterations that may not be observable ophthalmoscopically or detected with other examination techniques. (Posterior Segment Procedures, 1989).

FA is extremely valuable in studying the normal physiology of the retinal and choroidal circulation as well as demonstrating disease processes affecting the macula. (Clinical Ophthalmology, Kanski, 1989)

Fluorescein

Fluorescein is a stable, pharmacologically inert vegetable dye. Following intravenous injection, 80% of it binds to plasma proteins, mostly albumin. The remaining 20% is free and unbound within the bloodstream and is responsible for actual fluorescence during testing. When light energy of 465 to 490 nanometers (blue light) is directed at these fluorescein molecules, they fluoresce yellow-green with a peak emission of 520 to 530 nm. A fluorescein camera has a blue excitation filter (such as the Kodak Wratten 47) through which the camera flash passes. The resultant blue light continues into the patient's eye, exciting the intraocular free fluorescein to its fluorescing nanometer level. These fundus-reflected lights (blue and fluorescent yellow-green) return out of the eye through an introduced yellow-green filter barrier (such as Kodak Wratten G15). This filter absorbs the reflected blue light and allows only the emitted fluorescent light to be transmitted and recorded on high-speed black-and-white film in the camera.

Fluorescein's low molecular weight allows it to easily diffuse out of most of the body's capillaries, except for the normal vessels of the central nervous system, including the retinal vascular endothelium. This diffusion leads to the patient's skin becoming jaundiced in appearance for a few hours after testing and the urine becoming brilliant yellow for 24 to 48 hours. (Clinical Ophthalmology, Kanski, 1989)

Ocular Diseases Indicating the Need for Fluorescein Angiography

Acute posterior multifocal placoid pigment epitheliopathy (APMPPE)
Angiomatosis retinae
Anterior ischemic optic neuropathy
Bechet's disease
Branch retinal vein occlusion
Cavernous hemangioma of the retina
Choroidal rupture (when developing choroidal neovascularization)
Coat's disease (Retinal Telangiectasia)
Cystoid maculopathy (Irvine-Gass syndrome)
Diabetic retinopathy
Eale's disease
Fuch's spot (degenerative myopia)
Hemicentral retinal vein occlusion
Idiopathic central serous choroidopathy
Iris neovascularization
Maculopathy of angoid streaks
Malignant choroidal melanoma
Preretinal macular fibrosis
Presumed ocular histoplasmosis (macular changes)
Proliferative peripheral retinal disease
Retinal capillary hemangioma
Retinal macroaneurysm
Retinal pigment epithelial dystrophies (central)
Retinal pigment epithelial detachment
Retinal tumors
Sensory retinal detachments
Tumors of the iris and ciliary body

(Alexander, Larry, Primary Care of the Posterior Segment Disease, 1989.)

What to look for:

Areas of ***hypofluorescence*** caused by
1. Mechanical blockage from hemorrhage
2. Exudates
3. Glial tissue
4. Pigmentation
5. Vascular compromise from occlusion
6. Nonperfusion
7. Emboli
8. Arteriosclerosis

Areas of ***hyperfluorescence*** due to
1. Abnormal vasculature
   a. vessel tortuosity
   b. retinal or subretinal neovascularization.

Examples of abnormal angiograms:

A. Diabetic Retinopathy

1. Background Diabetic Retinopathy
   a. Microaneurysms hyperfluoresce
   b. Hemorrhages or exudates hypofluoresce

2. Macular Edema
   a. Fuzzy fluorescence or ground-glass appearance more noticeable in later phases indicates dye leakage into macula

3. Proliferative Diabetic Retinopathy
   a. Abnormally dark areas between retinal vessels indicate areas of capillary non-perfusion (hypoxia)
   b. Areas of neovascularization will fill with dye and be well-defined in earlier and middle phases. During later phases these areas will have a blurry or fuzzy appearance as the dye leaks out into the surrounding tissues

B. Occlusive Vascular Disease

1. Retinal Vein Occlusion (Branch and Central)
   a. Venous filling is delayed or does not occur in occluded area
   b. Overlying hemorrhages often obscure the view
   c. Collateral vessels, extravascular leakage and edema may also be noted

2. Retinal Artery Occlusion (Branch and Central)
   a. Arteriolar phase is delayed or does not occur in occluded area
   b. Venous phases are likewise affected
   c. Generally characterized by non-perfusion

C. Age-Related Maculopathy

1. Atrophic (Dry) ARMD
   a. Drusen hyperfluoresce
   b. Areas of pigment clumping hypofluoresce

2. Disciform (Wet) ARMD
   a. Sub-retinal (choroidal) neovascular membranes have a characteristic early filling during the choroidal phase
   b. Dye continues to leak during later phases

D. Aphakic Cystoid Macular Edema (Irvine- Gass Syndrome)

1. Macular hyperfluorescence occurs during later arteriovenous phases
2. Cystic spaces become well-defined during the late phase showing the characteristic "flower petal" pattern

E. Central Serous Retinopathy

1. Site of dye leakage usually begins as a small spot of hyperfluorescence in the early arteriovenus phase

2. Fluorescein continues to leak into the overlying sensory retinal detachment during later phases, often in a characteristic "smokestack" pattern

(Les Walls, Fluorescein Angiography, handout, 1990)

Potential side effects and complications:

1. Nausea
2. Vomiting
3. Vasovagal response
4. Discoloration of skin, urine, etc.
5. Pain and bruising at injection site
6. Allergic reactions
   a) mild - itching, rash
   b) **severe- bronchiospasm and anaphylactic shock**

Contraindications to performing a Fluorescein Angiogram

1. Renal failure
2. Allergy to fluorescein
3. History of surgery involving the lymphatics in the arm such as mastectomy (IV injection in that arm is contraindicated)
4. Pregnancy

(Les Walls, Fluorescein Angiography, handout, 1990)

Physiology of the Retina in Relation to the Free Circulating Fluorescein

Inner blood-retinal barrier:

The tight junctions of the retinal capillary endothelial cells form the inner blood-retinal barrier across which neither bound nor free fluorescein molecules can pass. Any leakage from the retinal circulation is therefore pathological.

Outer (RPE) blood-retinal barrier:

The major choroidal vessels are impermeable to both bound and free fluorescein molecules. On the other hand, the walls of the choriocapillaris are extremely thin and contain multiple fenestrations through which free (not bound) fluorescein molecules are able to escape into the extravascular space and also across Bruch's membrane creating a relatively uniform fluorescent background during testing referred to as the *choroidal flush*. However, adjacent cells of the RPE are firmly attached to each other by a series of adhesions called junctional complexes. These complexes (zonulae occludentes and zonulae adherentes) prevent the passage of free fluorescein molecules across the RPE and maintain the outer blood-retinal barrier. Passage of fluorescein across the RPE is therefore abnormal. (Clinical Ophthalmology, Kanski, 1989)

Circulation of fluorescein:

Fluorescein is injected into the median cubital vein and travels up the arm to the axillary vein and then to the subclavian where it enters the superior vena cava. Then, it travels into the right atrium to the right ventricle, where it proceeds to the pulmonary trunk, through the lungs and proceeds through the pulmonary veins to the left atrium. From that point, it procedes to the left ventricle where it is passed through the aorta for systemic circulation. Fluorescein then travels through either the brachiocephalic or left subclavian artery to reach the common carotid arteries where it enters the eye through the ophthalmic artery and then passes into the choroidal circulation through the short posterior ciliary arteries. From this point, it passes into the retinal circulation through the central retinal artery. Because the route to the retinal circulation is slightly longer than that to the choroidal circulation, the choroidal vessels are filled about 1 second before the retina. In the choroidal circulation often no precise details are discernible, mainly due to the rapid leakage of free fluorescein molecules from the choriocapillaris and also because the melanin in the RPE cells blocks choroidal fluorescence.

Phases of the Angiogram:

A. Anatomy

1. Choroid and choriocapillaris are like a leaky sponge
2. Normal RPE blocks view of underlying fluorescence
3. Macular pigment also blocks view somewhat

B. The normal fluorogram consists of four overlapping phases

Phase 1 is the pre-arterial phase which begins approximately 8-12 seconds after the dye injection, during which the choroidal circulation is filling, but no dye has reached the retinal arteries. This is referred to as the *choroidal flush.* The cilioretinal artery, if present, will fill during this stage.

Phase 2 is the arterial phase which follows 1 to 2 seconds after the pre-arterial phase. At this stage the central retinal artery and retinal arterioles fill. This stage extends from the first appearance of dye in the arteries until the whole arterial circulation is filled.

Phase 3 is the arteriovenous (capillary) phase, which is characterized by complete filling of the arteries and capillaries with early lamellar flow in the veins. Both arterioles and venules are uniformly filled as dye is being recirculated.

Phase 4 is the venous phase which can be further subdivided into early, mid and late stages according to the extent of venous filling and arterial emptying. Fluorescence of the arterioles begins to decrease as dye is being eliminated and the venules continue to fluorescence. In the late stage only faint residual staining of the choroid and retinal vessels remains in a normal eye.

Materials needed to perform fluorescein angiography:

1. T-max 400 Black and White film - 36 exposure
2. 5 ml of 10% fluorescein
3. Needle, filtration 19g x 1 1/2"
4. Needle, 23 gauge butterfly
5. 5 cc syringe
6. Alcohol swabs (2)
7. Small Bandage
8. Gloves
9. Blood pressure cuff/ sphygmomanometer/ stethoscope
10. Tourniquet - reusable
11. Emesis basin
12. Dilating drops

Les Walls, Fluorescein Angiography, handout, 1990

Routes of administration:

1. Intravenous (rapid sequence) fluorescein
2. Oral fluorescein angiography

Comparison of routes of administration:

a. Advantages of IV over oral
    1. Faster
    2. Better detail of choroidal and retinal vasculature (preferred technique prior to photocoagulation procedures)
    3. Able to detect choroidal neovascular nets and other anomalies visible during the early phases

b. Advantages of oral over IV
    1. Less likely to cause severe side effects than IV fluorescein angiography
    2. May be preferred technique for:
        a) Children
        b) Patients with inaccessible veins
        c) Patients who are very apprehensive about injections
    3. More likely to be performed legally by optometrists at the present time

RECIPE

"FLUORESCEIN FLUSH"
(as opposed to Orange Crush)

1. Patient should be fasted for 10-12 hours.
2. Take 1-2 grams of fluorescein solution (for injection) and place in non-transparent cup with non-transparent lid
3. Dilute with a small amount of chilled citrus beverage (Tang, diet pop, etc.)
4. Add small amount of crushed ice
5. Swirl to mix
6. Insert drinking straw, and have patient consume entire volume of fluorescein preparation within 1-2 minutes.
7. Examine fundus for evidence of late staining beginning at 10 to 15 minutes following ingestion
8. Examine fundus at 15 minute intervals for 60 minutes.

(Les Walls, Fluorescein Angiography, handout, 1990)

Technique for Performing IV (rapid sequence) Fluorescein Angiography

1. Thoroughly discuss the procedure with the patient. Make sure they understand why it is necessary and what the associated risks of the procedure are.
2. Have the patient sign an informed consent form, indicating their understanding.
3. Dilate pupils as appropriate for the conditions of the individual patient.
4. Check the crash cart and oxygen tank to make sure it is operating properly *(steroids, benadryl, epinephrine . . .).*
5. Fill the syringe from one ampule or vial of injectable 5%,10% or 25% sodium fluorescein (10% NaFl is the most often used).
6. Load one camera back with Kodachrome 64 and one with TMAX 400.
7. Take stereo photos or Polaroid photos of each eye on the color setting with the red-free filter and the patient's name plate (flash intensity of 18) for auto fluorescence.
8. REMOVE THE NAME PLATE (Otherwise the timer will not show).
9. Set the camera for fluorescein photos (flash intensity of 150-200) and take one filter test shot.
10. Prepare the patient for the injection of the fluorescein.
11. Fasten tourniquet around arm and find appropriate vein
    1. Antecubital area
    2. Back of the hand
12. Put on gloves.
13. Clean injection site with alcohol.
14. Have patient comfortably aligned in camera and set timer to zero.
15. Locate area of regard and adjust the fixation light appropriately.
16. Insert 23 gauge butterfly needle into vein and watch for blood to enter tubing.
17. Attach syringe with NaFl, remove tourniquet and start injecting fluorescein.
18. **Start timer** on camera and take the first photo.
19. The person making the injection, at 1 cc per second, calls out at mid-point and when the injection is completed.
20. When the injection is completed, a second photo is taken.
21. At the 12 second mark, 1 photo per second is taken for the next 10 seconds- by holding the camera button down, the camera will shoot every second.

22. At about the 25 second mark, take a photo every 2 seconds for five photos
23. At about the 40 second mark, take a photo every 5 seconds.
24. At 60 seconds, take a photo of the opposite eye for two shots.
25. At 70 seconds, take a photo every 5 seconds of the target eye.
26. At 120 seconds, take a photo of the opposite eye.
27. At 150 seconds, take a photo of the target eye and continue every 10 seconds.
28. At 180 seconds, take one shot every 30 seconds to one minute.
29. At 300 seconds, take two shots of the targeted eye and two of the opposite eye.
30. At 10 minutes take two more shots of each eye - if necessary.
31. Have the patient rest for twenty minutes following the procedure, again telling him/her about the discoloration of the skin and urine - if necessary.

(Les Walls, Fluorescein Angiography, handout, 1990)

Treatment of Anaphylaxis

1. Place patient in recumbent position and elevate lower extremities

2. Monitor vital signs often (every three minutes)

3. Apply tourniquet proximal to site of antigen injection: remove every 10 to 15 minutes.

4. Administer aqueous epinephrine 1:1000 into non occluded site
   Adults: 0.3 - 0.5 mL SC or IM
   Children: 0.01 mL/ kg, SC or IM q 15 min cm

5. Administer epinephrine 1:1000 into site of antigen injection
   Adults: 0.15 - 0.25 mL SC
   Children: 0.005 mL/kg, SC

6. Establish and maintain airway with cropharyngeal airway device, endotracheal intubation, transtracheal catherterization, or cricothyrotomy

7. Administer oxygen at 6-10 L/min *In office*

In E.R.

8. Institute rapid fluid replacement with saline or colloid solution

9. If hypertension is present
   Adults or children: dopamine (Dopastat, Intropin), 2:10 um/kg/min, or
   Adults: norepinephrine (Levophed), 8mg/500 mL dextrose (5%) in saline at 2mL/min and adjust rate to maintain low-normal blood pressure

10. If refractory hypotension is present:
    Cimetidine (tagamet), 300 mg, or ranitidine (Zantac), 50mg, IV, over 3 to 5 min

11. If bronchiospasm is present:
    Aminophyline, 6mg/kg, IV, over 20 min

12. To block late-phase reaction:
    Hydrocortisone sodium succinate (A-hydroCort, Solu-Cort), 100 mg, IV push, and 100 mg dextrose (5%) in saline, IV, q2-q4h

13. To block H, receptors:
    Diphenhydramine (Benadryl). 1-2 mg/kg, IV, up to 50 mg over 3 min

14. For adults taking a beta-adrenergic blocker
    Atropine, 0.5 mg, IV, q5min until heart rate is 60 beats/min, or Isoproterenol (Isuprel) drip 2-20 ug/min titrated to heart rate of 60 beats/min, or glucagon, 0.05 mg/kg bolus, IV, followed by 0.07 mg/kg/hr continuous IV infusion.

Les Walls, Fluorescein Angiography, handout, 1990

Reactions to Fluorescein and Treatment

Side Effects and Complications	*Treatment*
1. Nausea & Vomiting	1. Time and reassurance; have patient breathe through mouth slowly and deeply; reaction passes quickly 2. For those patients who have experienced nausea and vomiting from previous fluorescein injection, perform test in fasting state and give 25 to 50mg of Phenergan by mouth 1 hour before angiography
2. Fainting and syncopy (usually vasovagal; occurs more frequently in diabetic patients	1 Time and reassurance; further treatment is rarely necessary 2. Monitor pulse and blood pressure 3. Smelling salts 4. Oxygen 5. If severe, supine position and life-support measures.
3. Painful local extravasation	1. Ice pack to site of injection 2 Subcutaneous local anesthetic, rarely necessary
4. Generalized skin eruption (urticaria, pruitus)	1. Intravenous administration of antihistimine (Benadryl 25 to 50 mg)
5. Bronchiospasm and anaphylaxis (Acute medical emergency)	1. Provision of airway 2. Oxygenation 3. Close monitoring of vital signs 4. Systemic medications as needed a. Epinephrine b. Steroids c. Antihistamines d. Pressor agents; levarterenol (Levophed), metaraminol (Aramine) bitartrate e. Aminophylline

(Les Walls, Fluorescein Angiography, handout, 1990)

Management of Persons Exposed to Blood

Once an exposure has occurred, the blood of the individual from whom exposure occurred should be tested for hepatitis B surface antigen (HBsAg) and antibody to human immunodeficiency virus (HIV antibody). Local laws regarding consent for testing source individuals should be followed. Testing of the source individual should be done at a location where appropriate pretest counseling is available; post-test counseling and referral for treatment should be provided.

Human Immunodeficiency Virus Post-exposure Management

IF	THEN	AND
The source individual has AIDS OR The source individual is positive for HIV infection OR The source individual refuses to be tested	1. The exposed worker should be counseled about the risk of infection. 2. The exposed worker should be evaluated clinically and serologically for evidence of HIV infection as soon as possible after the exposure. 3. The exposed worker should be advised to report and seek medical evaluation for any febrile illness that occurs within 12 weeks after the exposure. 4. The exposed worker should be advised to refrain form blood donation and to use appropriate protection during sexual intercourse during the follow-up period, especially the first 6 - 12 weeks after exposure.	An exposed worker who tests negative initially should be retested 6 weeks, 12 weeks, and 6 months after exposure to determine whether transmission has occurred.
The source individual is tested and found seronegative	Baseline testing of the exposed worker with follow-up testing 12 weeks later may be performed if desired by the worker or recommended by the worker's health care provider.	
The source individual cannot be identified	Decisions regarding appropriate follow-up should be individualized. Serologic testing should be done if the worker is concerned that HIV transmission has occurred.	

1. Being "exposed to blood" means having blood, blood-contaminated saliva, or a blood-contaminated object come into contact with broken skin or mucous membranes, or pierce the skin as through a needlestick injury.

2. The information given in this table is based on recommendations in *Guidelines for Prevention of Transmission of HIV and HBV to Health-Care and Public-Safety Workers.* DHHS (NIOSH) Publication No. 89-107; Cincinnati, Ohio, February 1989.

Management of Persons Exposed to Hepatitis B

Once an exposure has occurred, the blood of the individual from whom exposure occurred should be tested for hepatitis B surface antigen (HBsAg) and antibody to human immunodeficiency virus (HIV antibody). Local laws regarding consent for testing source individuals should be followed. Testing of the source individual should be done at a location where appropriate pretest counseling is available; posttest counseling and referral for treatment should be provided.

IF	AND	THEN
The source individual is found positive for HBsAg	The exposed worker has not been vaccinated against hepatitis B.	1. The worker should receive the vaccine series for hepatitis B. 2. The worker should receive a single dose of hepatitis B immune globulin if it can be given within 7 days of exposure.
	The exposed worker has been vaccinated against hepatitis B.	The exposed worker should be tested for antibody to hepatitis B surface antigen (anti-HBs), and given one dose of vaccine and one dose of HBIG if the antibody level in the worker's blood sample is inadequate (i.e., <10 SRU by RIA, negative by EIA)
The source individual is found negative for HBsAg	The exposed worker has not been vaccinated against hepatitis B.	The worker should be encouraged to receive hepatitis B vaccine.
	The exposed worker has been vaccinated against hepatitis B.	No further action is needed.
The source individual refuses testing or cannot be identified	The exposed worker has not been vaccinated against hepatitis B.	1. The worker should receive the hepatitis B series. 2. HBIG administration should be considered on an individual basis when the source individual is known or suspected to be at high risk of HBV infection.
	The exposed worker has been vaccinated against hepatitis B.	Management and treatment of the exposed worker should be individualized.

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